

# Discovery

# Determination of sodium benzoate & potassium sorbate contamination in ready to serve products in post market surveillance in Chennai 2015

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#### **ABSTRACT**

Sodium benzoate and potassium sorbate are two major chemical preservatives which are used in ready to serve products. In this study, a total of 50 commercial brands of highly consumed ready to serve products were analysed. The HPLC determination of the preservatives was performed using a reversed-phase C<sub>18</sub>column and UV detection at 225 nm for sodium benzoate and 255 nm potassium sorbate. The preservative concentration in samples was quantified using authentic external standard sodium benzoate and potassium sorbate. Among 50 samples, the minimum and maximum concentration of benzoate content in various brands were 86 ppm and 674 ppm and for sorbate were 120ppm to 562 ppm respectively. 22% of samples do not compliance with FSSAI



standard, India. Exposure to these chemical preservatives could be a risk factor for the human health, especially when their intake was being occurred by various foodstuffs simultaneously. Stringent food regulation could be the best quick fix for the serious issue.

Keywords: Permissible preservatives, sodium benzoate, potassium sorbate, HPLC.

#### 1. INTRODUCTION

Chemical preservation has become an increasingly important practice in modern food technology and herbal medicinal products with the increase in production of processed food products (Mota FJM, et al., 2003). These preservatives are deliberately added to stop or delay nutritional and pharmaceutical product losses due to microbiological, enzymatic or chemical changes and thus increasing its shelf life and quality of foods. These also prevent consumer hazards due to the presence of microbial toxin or pathogenic microorganisms and economic losses due to spoilage. Benzoic acid (E210), sorbic acid (E200) and their corresponding salts are generally effective to control mold and inhibit yeast growth, and against a wide range of bacterial attack (Tfouni et al., 2001).

Although benzoic and sorbic acids and their salts are generally recognized as safe (GRAS) but the development of allergic reactions to benzoate in humans, such as uriticaria, non-immunological contact urticaria, metabolic acidosis, convulsions, hyperpnoea, weak clastogenic activity and asthma has been reported in some studies (Isabel MF et al., 2001). Other studies showed that sorbic acid has low toxicity, explained by the fact that it is rapidly metabolized by path ways similar to those of other fatty acids. In humans a few cases of idiosyncratic intolerance to sorbic acid and sorbate salts have been reported (non immunological contact urticaria and pseudo allergy). For these reasons, the use of food additives in different countries is limited by specific regulations. These preservatives are allowed by legislation but their use demands special care (Hajkova et al., 2002). The analytical determination of these preservatives is not only important for quality assurance purposes but also for consumer interest and protection. The most common analytical method for the determination of benzoic acid (BA) and sorbic acid (SA) or sodium benzoate (E211) and potassium sorbate (E202) is reversed-phase HPLC (B. Akbari et al., 2013). Here we report on a simplified procedure followed by HPLC separation of a mixture of sodium benzoate (E211) and potassium sorbate (E202). The purpose of this study was to quantify sodium benzoate and potassium sorbate in ready to serve products commercially available on the local markets in Chennai, Tamil Nadu in 2015.

#### 2. MATERIALS & METHODS

# Sample collection

The highly consumed ready to serve samples with different brand products were purchased from vendors in Chennai (Tamil Nadu). A total of 50 samples were collected to be representative of what a consumer would find in market-basket. Sample size ranged from 100 mL to1Litre and each sample was tested for the concentration of sodium benzoate and potassium sorbate. Out of 50 samples selected, 17 numbers of fruit products, 16 numbers of vegetable products and 17 number of carbonated water samples, in total 50 samples are randomly collected from the local market, Chennai (Tamil Nadu).

#### **Extraction & Cleanup**

25ml of ready to serve sample is taken after proper mixing, and transfer in to a 250 ml separatory funnel. 10 ml of hydrochloric acid (1:3) was added to the mixture. Benzoic acid & potassium sorbate was extracted thrice (as 50, 40, 30ml each) with diethyl ether. Combined ether extract with about 15 ml of water was washed by gentle swirling to remove any traces of mineral acid and discard aqueous phase. Ether layer was passed through anhydrous sodium sulphate and solvent on a water bath was removed and the last traces were removed by blowing air. Residue was dissolved in neutralised alcohol and titrated against 0.05 N NaOH solution using phenolphthalein as indicator. The titre (A) gives the titre equivalent to the mixture of benzoic acid /sorbic acid (AOAC, 2012).

# **Chromatographic conditions**

Purified extracts were analyzed by reversed-phase isocratic high performance liquid chromatography (HPLC) from Shimazu LC 10A using a Platinum C18 column (250  $\times$  4.6 mm id, 5  $\mu$ m) maintained at 40°C. A fluorescence detector was set at 375 nm (excitation) and 440 nm (emission). The mobile phase applied was deionized water/acetonitrile/methanol (60:20:20) with flow rate of 1.0 mL/min and injection volume of 20  $\mu$ L.

## Validation of analytical method

The analytical method was assessed for linearity, recovery, precision and limit of detection before sample analysis.

# 3. RESULTS & DISCUSSION

Fig. 1 illustrates the chromatogram of a standard solution, which contains 10mg/kg of sodium benzoate and 10mg/kg of potassium sorbate; the retention time of sodium benzoate and potassium sorbate are 9.176 and 9.548 min respectively. The elution order is potassium sorbate (retention time -9.176) and sodium benzoate (retention time - 9.548). Values found in the separation and the resolution of the column, indicate that the analytical method proposed in this work completely separates the analytes. The limit of detection (LOD) is defined as the smallest peak detected with a signal height three times that of the baseline while the limit of quantization (LOQ) referred to the lowest level of analyte which can be determined with an acceptable degree of confidence. Important analytical characteristics of the method are summarized in Table: 1. Peak identification of the preservatives in various samples was based on the comparison between the retention times of standard compounds and was confirmed by spiking known standard compounds to the sample. Quantification was based on the external standard method using calibration curves fitted by linear regression analysis. Chromatogram of both the preservative-positive sample is shown in figure 3 and 4. The average concentrations of sodium benzoate and potassium sorbate in ready to serve products were determined and given in Table 3 & 4.

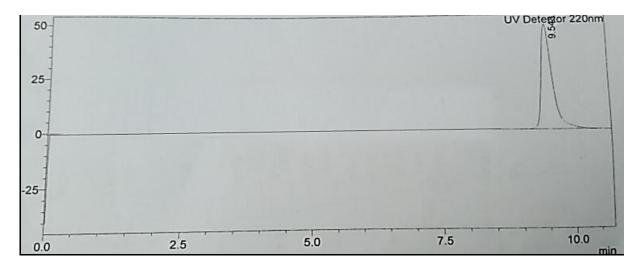


Figure 1 Illustration of standard sodium benzoate solution (10mg/kg)

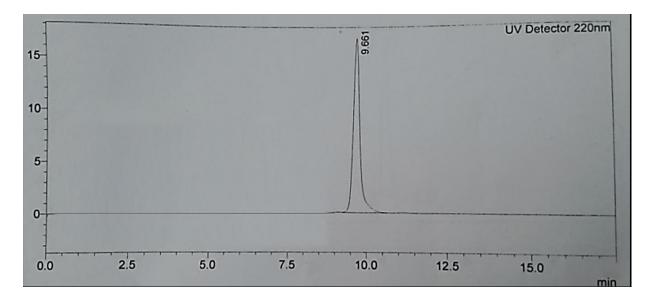


Figure 2 Illustration of standard Potassium sorbate solution (10mg/kg)

 Table 1
 Typical HPLC chromatogram of standard solutions of Sodium benzoate & potassium sorbate

| Peak name                    | Retention time | Area     | Height |
|------------------------------|----------------|----------|--------|
|                              | min            | mAU* min | mAU    |
| Standard -Sodium benzoate    | 9.548          | 712477   | 49185  |
| Standard - Potassium sorbate | 9.661          | 293196   | 22429  |

 Table 2 Analytical efficiency of HPLC technique

| Parameter                      | Preservative    |                   |
|--------------------------------|-----------------|-------------------|
|                                | Sodium benzoate | Potassium sorbate |
| Limit of detection (mg/l)      | 0.05            | 0.005             |
| Limit of quantification (mg/l) | 0.1             | 0.01              |

Table 3 Determination of Sodium benzoate concentration in ready to serve products

| Sample number | Sodium benzoate conc.<br>(mg/kg) | Permissible<br>limit(mg/kg) |
|---------------|----------------------------------|-----------------------------|
| 01            | 131                              | 120                         |
| 02            | 112                              | 120                         |
| 03            | 120                              | 120                         |
| 04            | 096                              | 120                         |
| 05            | 120                              | 120                         |
| 06            | 511                              | 600                         |
| 07            | 543                              | 600                         |
| 08            | 674                              | 600                         |
| 09            | 611                              | 600                         |
| 10            | 511                              | 600                         |
| 11            | 171                              | 120                         |
| 12            | 104                              | 120                         |
| 13            | 115                              | 120                         |
| 14            | 086                              | 120                         |

| 15 | 111 | 120 |
|----|-----|-----|
| 16 | 111 | 120 |
| 17 | 142 | 120 |
| 18 | 101 | 120 |
| 19 | 120 | 120 |
| 20 | 125 | 120 |
| 21 | 101 | 120 |
| 22 | 120 | 120 |
| 23 | 120 | 120 |
| 24 | 120 | 120 |
| 25 | 120 | 120 |

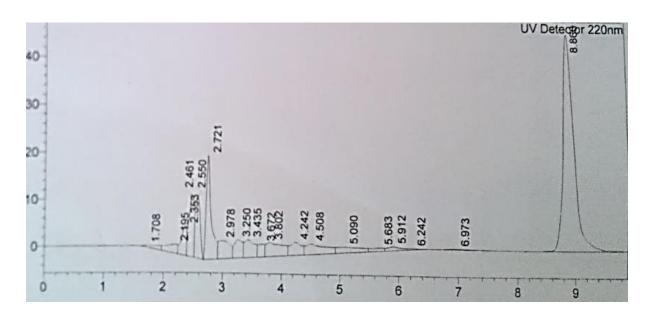


Figure 3 Illustration of sodium benzoate chromatogram of ready to serve product

FSSAI standard applauds sodium benzoate concentration in ready to serve fruit products should not exceed 120mg/kg, in ready to eat vegetable product it should not exceed 600mg/kg, in carbonated water it should not exceed 120mg/kg. 25 ready to serve samples are analysed for sodium benzoate concentration in it. Among them 24% of samples do not compliance with FSSAI standard i.e. exceeds the permissible limit.

**Table 4** Determination of Potassium sorbate concentration in ready to serve products

| Sample number | Potassium sorbate conc.(mg/kg)* |
|---------------|---------------------------------|
| 26            | 240                             |

| 27 | 562 |
|----|-----|
| 28 | 420 |
| 29 | 484 |
| 30 | 120 |
| 31 | 286 |
| 32 | 318 |
| 33 | 544 |
| 34 | 265 |
| 35 | 536 |
| 36 | 440 |
| 37 | 420 |
| 38 | 432 |
| 39 | 420 |
| 40 | 420 |
| 41 | 485 |
| 42 | 462 |
| 43 | 524 |
| 44 | 334 |
| 45 | 440 |
| 46 | 496 |
| 47 | 301 |
| 48 | 416 |
| 2  | 522 |
| 50 | 490 |

<sup>\*</sup>Allowable limit of potassium sorbate in RTS products: 500 mg/kg

FSSAI standard for potassium sorbate concentration in ready to serve fruit products, vegetable products and carbonated water should not exceed 500 mg/kg. 25 ready to serve samples were analysed using HPLC. Among them 20% of samples do not compliance with FSSAI standard i.e. exceeds the permissible limit.

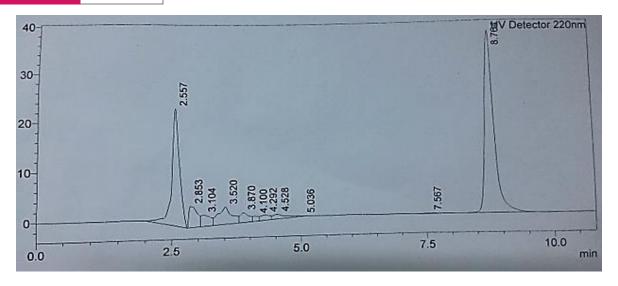


Figure 4 Illustration of Potassium sorbate chromatogram of ready to serve product

#### 4. CONCLUSION

Usage of these two permissible preservatives reduces the microbial bout in the sample and improves the shelf life of the products Akbari-adergani, S. Eskandari *et al.*, (2013). Recently studies reveals that these two preservatives do cause hazardous health effects like ADHD in children; irritation in eyes, nose, throat and skin; genotoxic effect in lympocytes; kidney, brain & nervous system disorder; mutagenic effects in the case of prolonged exposure Khosrokhavar et al., (2010). Therefore, the use of potassium sorbate and sodium benzoate should be regulated and more cooperation among producers, processors and the regional administration is essential.

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